

# **User Manual**

Version 2.0 Revision Date: 09/21/2015

Product name: BL21(DE3)pLysS Competent E. coli

Cat #: BP-100, BP-196

## **Description:**

- Chemically competent *E. coli* cells suitable for high yields, high efficiency transformation and protein expression.
- BL21(DE3)pLysS cells carry the lambda DE3 lysogen and pLysS plasmids.
- T7 Expression strain.

### **Application:**

- For expressing proteins in E. coli.
- Recommended for use when expressing toxic genes.

### Recommended storage condition:

This product should be stored at -80°C. Thaw on ice only before use. Do not re-freeze.

#### Genotype:

 $F^-$  ompT gal dcm(DE3) hsdSB(rB $^-$  mB $^-$ ) pLysS(Cam $^R$ )

#### **Protocol:**

- 1. Remove competent cells from -80°C and thaw competent cells on ice.
- 2. Add 5–10 ng of DNA to 50-100 μl of the cells and mix by tapping gently.
- 3. Incubate the cell on ice for 30 minutes.
- 4. Heat-shock the cells for exactly 30 seconds in the 42°C water bath.
- 5. Place the cells on ice for 2 min.
- 6. Add 250-500 µl of room temperature S.O.C medium to the cells
- 7. Shake at 225 rpm for 1 hour at 37°C.
- 8. Plate two different volumes of the transformation reaction onto LB plates containing the appropriate antibiotic for plasmid selection.
- 9. Incubate at 37°C overnight.

Note: Clones may exhibit differences in expression of heterologous genes. We recommend choosing 3–4 transformants when characterizing clones for protein expression.